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- (54) Polyacrylamide gel for medical and biological application and method of its preparation
- (57) A polyacrylamide gel for medical and biological application contains polyacrylamide and physiological solution with the following ratio of components (in % by weight):

polyacrylamide 3.0-28.0 physiological solution 72.0-97.0

The method of preparation of the said polyacrylamide gel comprises polymerization of acrylamide together with methylene-bis-acrylamide and elution of the final product, both polymerization and elution be-

ing carried out in the medium of the physiological solution. The polyacrylamide gel can be used as a base of nutrient mediums for growing microorganisms, artificial crystalline lenses and elastic contact lenses.

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# POLYACRYLAMIDE GEL FOR MEDICAL AND BIOLOGICAL APPLICATION AND METHOD OF ITS PREPARATION

Description of GB2114578

SPECIFICATION

Polyacrylamide gel for medical and biological purposes and process for preparing same

Technical Field

The invention relates to a polyacrylamide gel for medical and biological purposes, and to a process for preparing same. It is intended for use in medicine and biology.

Background Art

At present the natural agar gel is widely applied in microbiology. However, there exists a need to substitute natural materials for synthetic ones. Such a polymeric gel as a polyacrylamide gel is known for a comparatively long time and is used for various purposes due to its advantages.

In spite of this, the possibilities of its application for medical and biological purposes are restricted by the presence of toxic starting monomers within said gel. Known in the art is a synthetic growth medium for microorganisms (US Pat. No. 3,046,201, U.S. Cl. 195-100,

Published 24 July 1962) which medium contains 3 to 20 parts by weight of water per 1 part of a water soluble mixture of monomers which mixture in its turn contains 0.01 to 0.25 parts of one or another alkylidine-bis-acrylamide per 1 part of acrylamide.

The above polyacrylamide gel is a base whereto various mineral substances intended for nutrition of microorganisms are added. These substances may be introduced into the base at high concentrations to furnish the nutritive needs of certain types of microorganisms.

However, the above synthetic medium also contains a certain amount of toxic starting monomers. Besides, due to the fact that the synthetic base has an acid reaction (pH is 3.5 to 4).

some nutrient substrates can not be introduced into the base. The above consideration results in limitation of the possibility of use of said synthetic base.

A process for preparing a nutrient substrate for the cultivation of microorganisms (USSR inventor's Certificate No. 659,619,IPC2 C 12 K 1/06, Published 30.04:79) comprises preparing a polyacrylamide gel being a dense base for a nutrient substrate, followed by its impregnation with a nutrient substrate prior and after sterilization. The above process comprises polymerization of acrylamide and methylene-bis-acrylamide in an aqueous medium followed by its washing with water to remove the toxic starting monomers, prior to impregnating with a nutrient substrate. The base prepared in accordance with the above method is free of toxic nonreacted starting monomers thus widening the scope of applying this method in microbiology.

However, washing the end product with water providing for removal of the toxic substances therefrom, does not ensure the harmless contact of the resulting polyacrylamide gel with cells, tissues and organs of animals and human beings. The size of polyacrylamide gel being in contact with the living organisms is not stable.

Disclosure of Invention

The object of the present invention is to provide a polyacrylamide gel and a process for preparing same whose processing features will make it possible to ensure isosmotic properties of the polyacrylamide gel. and to widen its field of application.

The object set forth is attained by that the prior art polyacrylamide gel containing a polymer of acrylamide and of metylene-bis-acrylamide, according to the invention, further contains a physiological solution, the content of the components being the following (% by mass): polyacrylamide 3.0 to 28.0 physiological solution 72.0 to 97.0. The above polyacrylamide gel is non-toxic and features high porosity, hydrophily, elasticity, transparency, and thermal stability. In addition, the above polyacrylamide gel possesses isomotic properties to microorganisms, cells, tissues, and organs thus making it possible to stabilize the size thereof, and to increase its saturation with solutions of various substances. The above mentioned considerations allow a harmless contact between the polyacrylamide gel and the living organisms thus significantly widening the possibility of its application in medicine and biology.

In order to widen the possibility of application of the polyacrylamide gel in medicine and biology, it is expedient to use a 0.5% aqueous solution of sodium chloride, or a 0.9% aqueous solution of sodium chloride, or a Ringer-Lockart solution, or an Earle solution, or a Hanks solution, or an Eagle medium, or a 5% solution of glucose, as a physiological solution:

It is recommended that the polyacrylamide gel contain a 5% aqueous solution of sodium chloride as a physiological solution, the content of components being the following (% by mass): polyacrylamide 6.0 to 15.0 0.5% aqueous solution of sodium chloride 85.0 to 94.0

The above modification of the polyacrylamide gel makes it possible to obtain the most optimum isosmotic properties to microorganisms thus providing for its application as a dense base nutrient substrates intended for cultivating microorganisms:

It is possible for the polyacrylamide gel to contain a 0.9% aqueous solution of sodium chloride as a physiological solution; the content of the components being the following (% bymass): polyacrylamide 5:0 to 18:0 0.9% aqueous solution of sodium chloride 82:0 to 95.0

The above modification of the polyacrylamide gel makes it possible to obtain the most optimum isosmotic properties to animal and human cells thus providing for its application as a nutrient substrate carrier when cultivating cell cultures.

It is proposed that the polyacrylamide gel contain a 5% aqueous solution of glucose as a physiological solution, the content of the components being the following (% by mass): polyacrylamide 4.0 to 20:0 5.0% aqueous solution of glucose 80.0 to 96:0.

The above modification of the polyacrylamide gel provides for the optimum isosmotic properties to living organisms and is used in the instances when the presence of sodium chloride is undesirable.

The object set forth is also attained by that in the prior art process for preparing a polyacrylamide gel for medical and biological purposes which process comprises polymerizing acrylamide and methylene-bis-acrylamide followed by washing the end product, according to the invention, the steps of polymerizing acrylamide and methylene-bis-acrylamide, and of washing the end product are carried out in the physiological solution.

The process of the invention makes it possible to prepare a polyacrylamide gel which can be utilized as a dense base in growing essentially all the types of microorganisms, and also as an artificial crystalline lens, and a contact lens.

It is recommended to utilize a 0.5% aqueous solution of sodium chloride, or a 0.9% aqueous solution of sodium chloride, or a Ringer-Lockart solution, or an Earle solution, or a Hanks solution, or a 1 99 medium, or an Eagle medium, or a 5% aqueous solution of glucose, as a physiological solution.

The above modification of the process makes it possible to widen the possibility of utilization of the polyacrylamide gel in medicine and biology.

The polymerization process between acrylamide and methylene-bis-acrylamide is expedient to be carried out in a reactor whose inner cavity simulates the shape of an artificial crystalline lens with subsequent washing the end product, the processes of polymerization and washing being carried out within a medium of a 0.9% aqueous solution of sodium chloride.

The above modification of the process makes it possible to utilize a polyacrylamide gel as an artificial crystalline lens. In doing so, the traumatization of the eye tissues during the implantation is reduced due to carrying out minimum cuts and to providing for the complete areactivity of the eye tunics.

The polymerization process between acrylamide and methylene-bis-acrylamide is recommended to be carried out in a reactor whose inner cavity simulates the shape of a contact lens with subsequent washing the end product, the processes of polymerization and washing being carried out within a medium of a 0.9% aqueous solution of sodium

The above modification of the process makes it possible to utilize polyacrylamide gel as a contact lens. In doing so, there is provided a long-term continuous wearing when correcting refractive anomalies within a wide range.

Best Mode for Carrying Out the Invention

The polyacrylamide gel is prepared by polymerization of acrylamide and methylene-bisacrylamide or other water soluble alkylidine-bis-acrylamide. A reaction mixture generally contains 80 to 99.5% by mass of acrylamide, 0.5 to 20% by mass ofmethylenabis-acrylamide or any other water solublealkylidinebis-acrylamide. Starting monomers are dissolved within a physio logical solution. As a physiological solution there are used a 0.5% aqueous solution of sodium chloride, or a 0.9% aqueous solution of sodium chloride, or a 5% aqueous solution of glucose, or a Ringer-Lockart solution, or a Hanks solution, or an Earle solution, or an 99 medium, or an Eagle medium. By varying amounts of starting monomers in the reaction mixture, it is possible to obtain polyacrylamide gels of different density and elasticity.

The process of polymerization of the starting monomers can proceed with or without heating the reaction mixture, and with adding known initiators and catalysts. The polymerization rate is proportional to a temperature, to an amount of the catalyst, and to the intensity of irradiation.

The catalysts are usually added in amount of 0.05 to 0.1% based on the weight of the starting monomers.

A reaction mixture may be also prepared by mixing dry powdered monomers with their subsequent dissolution in a physiological solution. In order to speed up the process of dissolution, the physiological solution is heated. The polymerization of the reaction mixture may be carried out within a volume of a predetermined shape, i.e. in glass, metal, ceramic containers, and also in containers made from synthetic materials. The gel obtained as a result of the polymerization process assumes the shape and size of the utilized container.

The process of polymerization may be carried out within reactors whose cavity simulates the shape of the known contact lenses, or the shape of known monolithic artificial crystalline lenses, thus allowing the polyacrylamide gel to be utilized either as a soft contact lens; or as an artificial crystalline lens. The polymerization process conditions are similar to the above described. After the polymerization process is over the resulting polyacrylamide gel is washed with physiological solutions:

The process of washing the gel to remove starting monomers which have not entered into the polymerization reaction can be carried out under conventional conditions and at an increased temperature. In doing so, the starting monomers which have not entered into the reaction are dissolved in the physiological solution and go thereinto from the gel. By renewing the physiological solution, it is possible to completely remove toxic products from the gel. Usually for this purpose it is sufficient to renew the solution three times. Heating speeds up the process of washing the gel. The process of washing the gel is carried out similarly to the above described when obtaining a soft contact lens or a crystalline lens. The gel thus obtained is susceptible to impact extrusion and cutting. After washing and forming a required shape, the polyacrylamide gel is subjected to sterilization.

Sterilization of the gel can be accomplished in thermal, radiative, and chemical ways.

Selection of a sterilization method and its parameters are determined by the conditions of the specific task. After sterilization, the gel becomes suitable for the use as a dense base for preparing nutrient substrates for cultivating microorganisms, or as an artificial crystalline lens, or as a soft contact lens, and may be stored till its utilization:

Gel saturation with substrates intended for nutrition of microorganisms and cell cultures can be accomplished prior and after sterilization. Selection of a saturation method is determined by the composition of the nutrient substrate. With thermolabile components being present in the nutrient substrate, saturation is accomplished after sterilization. The composition of nutrient substrates is determined by nutritive needs of specific groups or kinds of microorganisms and cells. To saturate the polyacrylamide gel of the invention, nutrient substrates meeting nutritive needs of all the known kinds of microorganisms and cells including natural, semisynthetic; and synthetic substrate compositions or their mixtures can be utilized.

To determine the quality of nutrient media, and to examine biological properties of microorganisms, animal and human cells, conventional research methods are used.

Optical properties of soft contact lenses and artificial crystalline lenses made from a polyacrylamide containing a physiological solution are also determined by conventional methods.

The invention is further explained in terms of specific examples.

## Example 1

A polyacrylamide gel of the invention containing (% by mass)11% of polyacrylamide and 89% of a 0.5% aqueous solution chloride was prepared as follows. Three solutions (A, B, C) where previously prepared using the following procedure (amounts of starting components are given for 1000 ml of the basic solution): the solution A was prepared by dissolving 5 ml of tetramethyl ethylenediamine in 995 ml/of a 0.5% aqueous solution of sodium chloride and was stored in dark glassware at a temperature of C till the moment of utilization (conventional storage time is 6 to 8 months); the solution B was prepared by dissolving 7.35 g of methylenebis-acrylamide in 350 ml of a 0.5% aqueous solution of sodium chloride heated up to a temperature of 60°C. following which 280 g of acrylamide was added and mixed to a complete dissolving. The solution thus obtained was filtered through a cotton-gauze filter following which a 0.5% aqueous sodium solution was added up to an amount of 1000 ml, and the resulting solution was stored in dark glassware within a refrigerator at a temperature of 4°C till the moment of utilization (conventional storage time is 6 to 8 months); the solution C was prepared by dissolving 1.4 9 of ammonium persulfate in 1000 ml of a 0.5% aqueous solution of sodium chloride and was stored in dark glassware till the moment of utilization (conventional storage time is 4 to 6 weeks).

A reaction mixture was prepared from the solutions A, B, C. For this purpose 2 volumetric parts of the solution B, and 4 volumetric parts of the solution C were added to 1 volumetric part of the solution A.

The reaction mixture was poured into a slot formed by two plane parallel glass plates each having a thickness of 3 mm. The polymerization process proceeded for 1.5 minutes. The glass plates were then separated and a plate of the polyacrylamide gel thus formed was released.

Circular disks being 70 mm in diameter were pressed out of the gel plate. The disks thus obtained were placed into a container and were poured over with a 0.5% aqueous solution of dium chloride (20 ml of the solution per 1 disk). The disks were then exposed in a 0.5% aqueous solution of sodium chloride for 1.2 hours with renewing the solution after every 4 hours. After 12 hours the solution was poured off.

The resulting polyacrylamide gel is non-toxic, possesses high porosity, hydrophily, elasticity, transparency, and thermal stability.

Cells of various kinds of microorganisms applied to the gel preserved their shape, size and viability during a long time (3 months or more) thus indicating isosmotic properties of the given gel to the cells of microorganisms.

The gel was readily saturated with such substrates for nutrition of the microorganisms as a beef-extract broth, therefore it was utilized as a dense base for preparing nutrient substrates intended for cultivation of various groups of microorganisms (Escherichia, Salmonella, Shigella, Proteus, Staphylococcus etc.):

For the purpose of saturation, the washed disks were poured over with a Hottinger broth and amine nitrogen in an

amount of 300 mg % (10 ml of broth per 1 disk) and were subjected to sterilization at a temperature of 120 for 30. minutes. During this time saturation with the broth and sterilization were carried out. Sterile disks saturated with the broth, kept under sterile conditions, where placed into sterile Retri dishes, were dried and seeded with Escherichia

The microorganisms were subjected to incubation at a temperature of 37°C for 24 hours. During this time the culture of Escherichia coli in the form of colonies grew on the disk surfaces.

During their growth on the gel, the microorganisms preserved such biological properties as nature of growth and fission, shape of cells and colonies, morphological, tinctorial, cultural, biochemical, serological properties, antigenic structure, phagolysability. The biomass of the grown microorganisms, with the seeding dose being the same, exceeded the biomass of similar microorganisms grown on the beef-extract agar.

The survival of investigated types of microorganisms on the polyacrylamide gel of the invention and on the prior art gel was also determined. The results are given in Table 1.

### Table 1

Average amount

of grown co- Average amo

lonies unt of grown-

(polyacryl- colonies

Seeding does of amide gel (prior art

Kind and strain of colony forming of the in- nutrient microorganisms units vention) substrate)

Staphylococcus 100 94 82 aureus 209P

E. coli M-17 100 96 80E.coli K-12 100 92 84

B.cereus 8035 100 88 72

B:mesentericus 1027 100 94 82

B.subtilis 83 100 92 76

B.megaterium 654 100 89 81

Sh.sonnei 100 76 71

Sh.flexneri 100 84 70

Pseudomonas aeruginosa 165 100 92 86

Example 2

A polyacrylamide gel of the invention containing (% by mass) 8.0% of polyacrylamide and 92% of a physiological solution was prepared in accordance with the method specified in

Example 1. A 0.9% aqueous solution of sodium chloride was used as the physiological solution.

The resulting polyacrylamide gel was non-toxic; possessed high porosity, hydrophily, elasticity, transparency, and thermal stability. The gel was readily saturated with substrates for nutrition of microorganisms, animal and human

A 2% suspension of sheep erythrocytes applied to the gel surface in an amount of 0.2 ml was not subjected to hemolysis, and fibroblasts, HeLa and KB cells applied thereto preserved their shape, size, and viability for up to two days at a temperature of 4°C. Intraperitoneal implantation of plates of the obtained get sizing 1 cm x 1 cm x 0.3 cm to white mice has not indicated any changes of initial plate size for five days. The plates remained transparent, did not cause reactive changes of surrounding organs and tissues, and were well endured by animals even in the case of simultaneous implantation of 2 to 3 plates to one animal.

A polyacrylamide gel of the invention containing 3.0% by mass of polyacrylamide and 97.0% by mass of a physiological solution was prepared in accordance with the method specified in

Example 1. A Ringer-Lockart solution was used as the physiological solution.

The properties of the resulting polyacrylamide gel were similar to those described in Example 2.

Shape, size and viability of fibroblasts, HeLa and KB cells were preserved for up to 4 days at a temperature of C

A polyacrylamide gel of the invention containing 11.0% by mass of polyacrylamide and 89.0% by mass of a physiological solution was prepared in accordance with the method specified in Example 1. A Hanks solution was used as the physiological solution.

The properties of the resulting polyacrylamide gel were similar to those described in Example 2.

Plates of said gel were rose-colored, and shape, size, and viability of fibroblasts, HeLa and KB cells remained on said gel for up to 6 days at a temperature of4 C.

The gel plates thus obtained were saturated with a growth medium for cell cultures which medium contained 60% of al 99 medium, 20% of lactalbuminum hydrolysate, 20% of cattle serum, and were used for growing HeLa cells. The HeLa cells grew on the gel surface in the form of a typical monolayer during conventional terms.

## Example 5

A polyacrylamide gel of the invention containing 20.0% by mass of polyacrylamide and 80.0% by mass of a physiological solution was prepared in accordance with the method specified in Example 1.

An Earle solution was used as the physilogical solution.

The properties of the resulting polyacrylamide gel were similar to those described in Examples 2 and 4

#### Example 6

A polyacrylamide gel of the invention containing 5.0% by mass of polyacrylamide and 95.0% by mass of a physiological solution was prepared in accordance with the method specified in Example 1.\*

A 199 medium was used as the physiological solution.

The properties of the resulting polyacrylamide gel were similar to those described in Examples 2 and 4

Shape, size, and viability of fibroblasts, HeLa and KB cells were preserved on the above gel for up to 8 to 10 days at a temperature of4 C.

The cells grown on the gel plates were well fixed thereto thus making it possible to transfer gel blocks with cells which blocks were cut out of said plates; and to carry out microscopic investigation thereof, and also to place blocks with cells into microchambers.

Example 7

A polyacrylamide gel of the invention containing 15.0% by mass of polyacrylamide and 85.0% by mass of a physiological solution was prepared in accordance with the method specified in Example 1.

A1 99 medium was used as the physiological solution.

The properties of the resulting polyacrylamide gel were similar to those described in Examples 2 and 4. A monolayer of cells grown on the gel plates was readily washed off thus making it possible to accumulate the cell biomass.

Example 8

A polyacrylamide gel of the invention containing 7.0% by mass of polyacrylamide and 93% by mass of a physiological solution was prepared in accordance with the method specified in Example 1.

An Eagle medium was used as the physiological solution.

The properties of the resulting polyacrylamide gel were similar to those described in Examples 2 and 7.

The gel plates saturated with a growth medium were used in growing cell strains. In doing so, a good growth of diploid cells was observed.

Example 9

A polyacrylamide gel of the invention containing (% by mass) 10.0% of polyacrylamide and 90.0% of a physiological solution was prepared in accordance with the method specified in

Example 1: A 5% aqueous solution of glucose was used as the physiological solution.

The properties of the resulting polyacrylamide gel were similar to those described in Example 2.

In addition, shape size, an viability of fibroblasts, HeLa and KB cells were preserved for up to 3 days at a temperature of4.C.

At the same time, there were noted an acceleration in growth and an increase in the biomass of microorganisms containing saccharolytic enzymes. The fact that gel composition was free from sodium chloride resulted in a considerable simplification of determining the amount of sodium chloride in the microorganism cells.

Example 10

A polyacrylamide gel of the invention containing (% by mass) 11.0% of polyacrylamide and 89.0% of a 0.9% aqueous solution of sodium chloride was prepared as follows. Three basic solutions (A, B, C) were previously prepared. Given below are the amounts of starting components calculated for 1 l of each solution. The solution A was prepared by dissolving 5 ml of tetramethyl ethylenediamine in 995 ml of a 0.9% aqueous solution of sodium chloride. The solution B was prepared by dissolving 7.35 ml of methylne-bis-acrylamide in 350 ml of a 0.9% aqueous solution of sodium chloride heated up to a temperature of 60°C, following which 280 g of acrylamide were added, the resulting mixture being mixed, filtered, and a 0.9% aqueous solution of sodium chloride was added up to an amount of 1000 ml. The solution C was prepared by dissolving 1.4 g of ammonium persulfate in 1000 ml of a 0.9% aqueous solution of sodium chloride.

A reaction mixture was prepared from the above basic solutions. The solutions A, B, C were taken in a volumetric ratio therebetween of 1:2:4.

Th volumetric ratio between the basic solutions in preparing the working mixture may be changed depending on a required degree of elasticity of an artificial crystalline lens.

The reaction mixture thus prepared was poured in an amount of 0.5 ml into a reactor whose cavity simulated both the

optical and the supporting portions of the crystalline lens. The reaction mixture was polymerized for 3 minutes at a temperature of 20°C. After the given time interval had been over, the artificial crystalline lens thus obtained was released from the reactor. The artificial crystalline lens is characterized by the following parameters: the curvature radius of the front surface is 27, 22 mm, the rear surface is plane; the diameter of the optical portion is 6 mm, the refraction index is + 18.0 D.

After removal, the crystalline lens was washed in 20 ml of a 0.9% aqueous solution of sodium chloride for 24 hours with renewing the solution three times. The crystalline lens was sterilized in a 0.9% aqueous solution of sodium chloride for 40 minutes by boiling and was stored in the same solution within a sealed vessel till utilization.

On an aphacic eye with preserved rear capsule of the crystalline lens, a cut was made in a comea scleral or a corneal zone, said cut having a length of up to 4.5 mm. The rolled crystalline lens was inserted with forceps into the formed. opening and was moved through the coloborna or through the pupil into the posterior chamber. After releasing the forceps branches the crystalline lens straightened out due to its elasticity, the supporting portions rested upon the capsule equator thus promoting lens alignment and its secure fixation caused by resilient properties of the supporting portions. The use of the inventive crystalline lens is possible either in application of conventional methods of one-time extracapsular extraction of cataract or as a second stage, and in application of phacoemulsification

## Prior art artific

ial crystalline

Inventive artificial lens made from poly

Parameter crystalline lens methylmethacrylate

Cut length, mm 4.5 6

During the postoperative period, a moderate injection of the eyeball was noted, said injection being commensurable with the control. Abatement of inflammation symtoms in active application of antibiotics, hormone drugs, and mydriatics took place during a period of 3 to 4 weeks.

The results of the carried out operative procedures were traced for 24 months. The following features were noted: absence of symptoms of the chronic inflammation, secure fixation of the implanted body, stable refraction ability.

A polyacrylamide gel of the invention containing (% by mass) 5:0% of polyacrylamide and 95:0% of a physiological solution was prepared in accordance with the method specified in

Example 1 0. A 0.9% aqueous solution of sodium chloride was used as the physiological solution. The polymerization process was carried out in a reactor whose inner cavity simulated the shape of the prior art contact lens.

A soft contact lens was obtained, said lens having the following parameters: the curvature radius of the front surface is 8.2 mm, the curvature radius of the rear surface is 7.7 mm, the diameter is 14.5 mm, the refraction is 1.75 D.

In experiments carried out on animals (10 rabbits), soft contact lenses were placed onto the cornea for a long-term continuous wearing during 2 to 4 weeks.

The following parameters were checked: displaceability, condition of the corneal epithelium checked by biomicroscopy with a fluorescein, response of the eyeball conjunctiva and of the eyelids. A moderate displaceability of lenses was noted, said displaceability being not more than 1 mm, while such phenomena as allergization or imitation of the conjunctiva, edema or erosion of the corneal epithelium were not detected.

In experiments carried out on the authors and consisting in a long-term continuous wearing of the soft contact lenses (for 4 to 6 weeks) there were noted a rapid getting accustomed to said lenses, and the absence of troublesome feedlings. The complete areactivity of the eyeball conunctive and the eyelids was determined, and such phenomena as edema or erosion of the corneal epithelium were not detected. The displaceability of the soft contact lenses did not exceed 1 mm.

Example 12

A polyacrylamide gel of the invention containing (% by mass) 5.0% of polyacrylamide and 95.0% of a physiological solution was prepared in accordance with the method specified in

Example 10. A 0.9% aqueous solution of sodium chloride was used as the physiological solution. The polymerization process 71.0% by mass of a physiological solution was prepared according to the method specified in

A 0.9% aqueous solution of sodium chloride was used as the physiological solution. The polyacrylamide gel thus obtained was of a very dense consistence and exhibited brittleness when bent. Washing the gel plates to remove the starting components was not effective and required a long time interval (15 days or more). The gel was inadequately saturated with substrates for the nutrition of microorganisms and cell cultures, was fast-drying with crack initiation at a temperature of 37'C, and was difficult to fix in Petri cups.

INDUSTRIAL APPLICABILITY

The polyacrylamide gel can be used as a dense base for applying nutrient substrates that are necessary for growth, fission and development of microorganisms.

Utilization of the polyacrylamide gel as a dense base of nutrient media ensures microbiological inertia of the latter thereby increasing the output of the microorganism biomass 1:5-2 times.

As a synthetic preparation, the polyacrylamide get has a conventional and fixed composition ensuring reproducibility of

the dense base of nutrient media and standardization of microbiological tests, which allows the results of different laboratories to be compared.

The plates of the polyacrylamide gel may be shaped into circles according to the diameter of ...
Petry dishes, into squares or rectangles according to the dimensions of cover glasses or slides and also into blocks of different dimensions and shapes, and after saturation with nutrient substrates, they can be utilized for macro and microcultivation of different groups, species and strains of microorganisms, cells of animals and man.

Production of the polyacrylamide gel can be automated.

The plates of the gel do not require special method of sterilization. Their sterilization may be carried out by conventional methods at usual condition.

The ready-to-use plates of the polyacrylamide gel may be stored for a long time.

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# POLYACRYLAMIDE GEL FOR MEDICAL AND BIOLOGICAL APPLICATION AND METHOD OF ITS PREPARATION

Claims of GB2114578

### CLAIMS

1. A polyacrylamide gel for medical and biological purposes containing a polymer of acrylamide and of methylene-bisacrylamide: characterized in that the polyacrylamide gel further contains a physiological solution, the content of the components being (% by mass): polyacrylamide 3.0 to 28.0 physiological solution 72.0 to 97.0

2. A polyacrylamide gel for medical and biological purposes according to claim 1, characterized in that the physiological solution is a 0.5% aqueous solution of sodium chloride; or a 0.9% aqueous solution of sodium chloride; or a Ringer-Lockart solution, or an Earle solution, or a

Hanks solution, or a medium 199, or an Eagle medium, or a 5% aqueous solution of glucose.

3. A polyacrylamide gel for medical and biological purposes according to claim 2, characterized in that it contains a 0.5% aqueous solution of sodium chloride as the physiological solution, the content of the components being (% by mass): polyacrylamide 6.0 to 15.0 0.5% aqueous solution of sodium chloride 85.0 to 94.0

4. A polyacrylamide gel for medical and biological purposes according to claim 2, characterized in that it contains a 0.9% aqueous solution of sodium chloride as the physiological solution, the content of the components being (% by mass): polyacrylamide 5.0 to 18.0 0.9% aqueous solution of sodium chloride 82.0 to 95.0

5. A polyacrylamide gel for medical and biological purposes according to claim 2, characterized in that it contains a 5% aqueous solution of glucose as the physiological solution, the content of the components being (% by mass): polyacrylamide4:0 to 20.0.5% aqueous solution of glucose 80.0 to 96:0

6. A process for preparing a polyacrylamide gel for medical and biological purposes according to claim 1 comprising polymerizing acrylamide andmethylene-bis-acrylamide followed by washing the end product, characterized in that polymerizing acrylamide andmethylene-bis- acrylamide, and washing the end product are carried out in the medium of a physiological solution.

- 7. A process for preparing a polyacrylamide gel for medical and biological purposes according to claim 6, characterized in that the physiological solution is a 0.5% aqueous solution of sodium chloride, or a 0.9% aqueous solution of sodium chloride, or a Ringer-Lockart solution, or an Earle solution, or a Hanks solution, or a medium 199, or an Eagle medium, or a 5% aqueous solution of glucose.
- 8: A process for preparing a polyacrylamide gel for medical and biological purposes according to claim 6, characterized in that polymerizing acrylamide and methylene-bis-acrylamide is carried out in a reactor whose inner cavity simulates the shape of an artificial crystalline lens with subsequent washing the end product, polymerizing and washing being carried out in the medium of a 0.9% aqueous solution of sodium chloride.
- 9. A process for preparing a polyacrylamide gel for medical and biological purposes according to claim 6, characterized in that polymerizing acrylamide and methylene-bis-acrylamide is carried out in a reactor whose inner cavity simulates the shape of a contact lens with subsequent washing the end product, polymerizing and washing being carried out in the medium of a 0.9% aqueous solution of sodium chloride.

# AMENDED CLAIMS OF INTERNATIONAL APPLICATION PCT/SU 80/00104

1. A polyacrylamide gel for medical and biological purposes containing a copolymer of acrylamide and of methylene-bis-acrylamide, characterized in that the polyacrylamide gel further contains a physiological solution, the content of the component being (% by mass): polyacrylamide 3.0 to 28:0 physiological solution 72.0 to 97.0 2. A poliacrylamide gel for medical and biological purposes according to claim 1, characterized in that the physiological solution is a 0:5% aqueous solution of sodium chloride; or a 0:9% aqueous solution of sodium chloride, or a Ringer-Locart solution, or an Earle solution, or a Hanks solution, or a medium 199, or an Eagle medium, or a 5% aqueous solution of glucose.

- 3. A polyacrylamide gel for medical and biological purposes according to claim 2, characterized in that it contains a 0.5% aqueous solution of sodium chloride as the physiological solution, the content of the components being (% by mass): polyacrylamide 6.0 to 15.0 0:5% aqueous solution of sodium chloride 85.0 to 94.0
- 4. A polyacrylamide gel for medical and biological purposes according to claim 2, characterized in that it contains a 0.9% aqueous solution of sodium chloride as the physiological solution, the content of the components being (% by mass): polyacrylamide 5.0 to 18.0 0.9% aqueous solution of sodium chloride 82.0 to 95.0
- 5. A polyacrylamide gel for medical and biological purposes according to claim 2; characterizzed in that it contains a 5% aqueous solution of glucose as the physiological solution, the content of the components being (% by mass); polyacrylamide 4.0 to 20.0.5% aqueous solution of glucose 80.0 to 96.0
- 6. A process for preparing a polyacrylamide gel for medical and biological purposes according to claim 1 comprising copolymerizing acrylamide and methylene-bis-acrylamide followed by washing the end product, characterized in that copolymerizing acrylamide and methylene-bis-acrylamide and washing the end product are carried out in the medium of a physiological solution.
- 7. A process for preparing a polyacrylamide gel for medical and biological purposes according to claim 6, characterized in that the physiological solution is a 0.5% aqueous solution of sodium chloride, or a 0.9% aqueous solution of sodium chloride, or a Ringer-Lockart solution, or an Earle solution, or a Hanks solution, or a medium 199, or an Eagle medium, or a 5% aqueous solution of glucose.

8. A process for preparing a polyacrylamide gel for medical and biological purposes according to claim 6; characterized in that copolymerizing acrylamide and methylene-bis acrylamide is carried out in a reactor whose inner cavity simulates the shape of an artificial crystalline lens with subsequent washing the end product, copolymerizing and washing being carried out in the medium of a 0.9% aqueous solution of sodium chloride.

9. A process for preparing a polyacrylamide gel for medical and biological purposes according to claim 6, characterized in that copolymerizing acrylamide and methylene-bisacrylamide is carried out in a reactor whose inner cavity simulates the shape of a contact lens with subsequent washing the end product, copolymerizing and washing being carried out in the medium of a 0.9% aqueous solution of sodium chloride.

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